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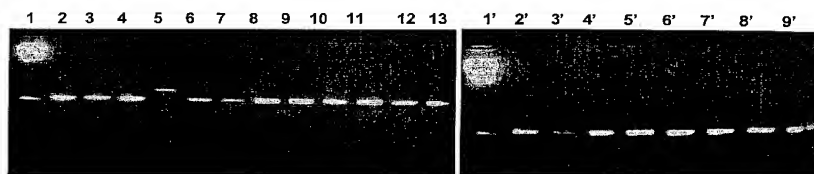
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[Continued on next page]

(54) Title: ASSAY FOR DETECTING AND IDENTIFYING MICRO-ORGANISMS

Amplification of molecular marker I (pur A) in Gram-positive bacteria



1 = DNA Ladder ( $\lambda$ /Hind III)  
2 : *Streptococcus pyogenes*  
3. *Streptococcus pneumoniae*  
4. *Streptococcus oralis*  
5. *Enterococcus hirae*  
6. *Enterococcus casseliflavus*  
7. *Streptococcus agalactiae*  
8. *Streptococcus sanguis*  
9. *Enterococcus faecalis*  
10. *Enterococcus gallinarum*  
11. *Enterococcus faecium*  
12. *Enterococcus flavescens*  
13. *Enterococcus durans*

1' : DNA Ladder ( $\lambda$ /Hind III)  
2' : *Enterococcus raffinosus*  
3' : *Enterococcus villorum*  
4' : *Staphylococcus aureus*  
5' : *Staph. epidermidis*  
6' : *Staphylococcus hominis*  
7' : *Bacillus anthracis*  
8' : *Bacillus cereus*  
9' : *Bacillus megatherium*

(57) Abstract: The present invention also relates to an assay for detecting and identifying micro-organisms, and in particular bac-  
teria. The present invention also relates to an assay for detecting micro-organisms, and in particular bacteria, in a sample, and for  
the discrimination thereof. More in particular the present invention relates to an assay for the molecular identification of bacteria  
according to Gram-, genus- species- and strain-specificity based on multigenotypic testing of bacterial DNA from human, animal or  
environmental samples.

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SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

— *before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments*

**Declaration under Rule 4.17:**

— *of inventorship (Rule 4.17(iv)) for US only*

**(88) Date of publication of the international search report:**

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— *with international search report*

*For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.*

## INTERNATIONAL SEARCH REPORT

International Application No  
PCT/EP2005/002927

**A. CLASSIFICATION OF SUBJECT MATTER**

IPC 7 C12Q1/68

According to International Patent Classification (IPC) or to both national classification and IPC

**B. FIELDS SEARCHED**

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 C12Q

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, BIOSIS, EMBASE, WPI Data, PAJ

**C. DOCUMENTS CONSIDERED TO BE RELEVANT**

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	CARROLL N M ET AL: "Detection of and discrimination between gram-positive and gram-negative bacteria in intraocular samples by using nested PCR" JOURNAL OF CLINICAL MICROBIOLOGY, WASHINGTON, DC, US, vol. 38, no. 5, May 2000 (2000-05), pages 1753-1757, XP002308232 ISSN: 0095-1137 abstract page 1754, column 1, paragraph 5; figure 1; table 2  ----- -/--	1-3,8

☒ Further documents are listed in the continuation of box C.☒ Patent family members are listed in annex.

## ° Special categories of cited documents:

- \*A\* document defining the general state of the art which is not considered to be of particular relevance
- \*E\* earlier document but published on or after the international filing date
- \*L\* document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- \*O\* document referring to an oral disclosure, use, exhibition or other means
- \*P\* document published prior to the international filing date but later than the priority date claimed

- \*T\* later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
- \*X\* document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
- \*Y\* document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.
- \*G\* document member of the same patent family

Date of the actual completion of the international search

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Name and mailing address of the ISA

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## C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	<p>KLASCHIK SVEN ET AL: "Real-time PCR for detection and differentiation of gram-positive and gram-negative bacteria." JOURNAL OF CLINICAL MICROBIOLOGY. NOV 2002, vol. 40, no. 11, November 2002 (2002-11), pages 4304-4307, XP002334305 ISSN: 0095-1137 cited in the application abstract page 4304, column 1, paragraph 4 - column 2, paragraph 4 page 4305, column 1, paragraph 3 - column 2 page 4304, column 1, paragraph 3 - paragraph 4 page 4304, column 2, paragraph 3 - paragraph 4 page 4305, column 2, paragraph 2</p>	1-3,8
X	<p>WO 90/11370 A (HOLLAND BIOTECHNOLOGY BV) 4 October 1990 (1990-10-04) abstract page 3, paragraph 2 - page 8, paragraph 1; claims 1-8</p>	1,2,8
X	<p>US 6 617 156 B1 (DOUCETTE-STAMM LYNN A ET AL) 9 September 2003 (2003-09-09) abstract column 19, paragraph 3 - column 20, paragraph 1 column 25, paragraph 4 - paragraph 6 contig 547: SEQ ID NO 1419: purA gene column 27, paragraph 2 - paragraph 3; tables 2,3</p>	9,11, 13-16
X	<p>LEEUVEN VAN W B ET AL: "Multilocus sequence typing of Staphylococcus aureus with DNA array technology" JOURNAL OF CLINICAL MICROBIOLOGY, WASHINGTON, DC, US, vol. 41, no. 7, July 2003 (2003-07), pages 3323-3326, XP002280375 ISSN: 0095-1137 abstract page 3323, column 1, paragraph 3 - page 3324, column 1, paragraph 1 page 3325, column 1, paragraph 2 - column 2, paragraph 2; tables 1-3</p>	1,2,8
X	<p>WO 98/20160 A (E.I. DU PONT DE NEMOURS AND COMPANY; HAZEL, JAMES, WILLIAM; JENSEN, MA) 14 May 1998 (1998-05-14) the whole document</p>	1,2,8

## INTERNATIONAL SEARCH REPORT

International application No.  
PCT/EP2005/002927

### Box II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:  
because they relate to subject matter not required to be searched by this Authority, namely:
2. ☐ Claims Nos.:  
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
3. ☐ Claims Nos.:  
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

### Box III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☒ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

1-5, 8, 9, 11, 13-16 (in part)

#### Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

## FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

Invention 1: claims 1-5,8,9,11,13-16 (in part)

Assay, its use, a primer pair, a nucleic acid probe, a composition, a kit and a DNA chip for detecting and identifying one or more microorganisms in a sample involving the molecular marker PurA (marker I) as identified in Table 1 of the application

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Invention 2-4: claims 1-5,8,9,11,13-16 (in part)

Assay, its use, a primer pair, a nucleic acid probe, a composition, a kit and a DNA chip for detecting and identifying one or more microorganisms in a sample involving the molecular marker identified by markers II-IV as identified in Table 1

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Invention 5-8: claims 1-3,6-8,10,12-16 (in part)

Assay, its use, a primer pair, a nucleic acid probe, a composition, a kit and a DNA chip for detecting and identifying one or more microorganisms in a sample involving the molecular marker identified by markers V-VIII as identified in Table 1

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## INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/EP2005/002927

Patent document cited in search report		Publication date	Patent family member(s)	Publication date
WO 9011370	A	04-10-1990	AU 5351790 A EP 0464123 A1 JP 5504672 T NL 8900670 A	22-10-1990 08-01-1992 22-07-1993 16-10-1990
US 6617156	B1	09-09-2003	NONE	
WO 9820160	A	14-05-1998	AU 5100798 A DE 69722028 D1 DE 69722028 T2 EP 0948643 A1	29-05-1998 18-06-2003 18-03-2004 13-10-1999